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Surface-Bound Membrane-Mimetic Assemblies: Electrostatic Attributes of Integral Membrane Proteins.

12 PERSONAL AUTHOR(S)
H. Gilbert Smith, Ph.D.

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The objective of this project was to develop a system for investigating stimulus-induced displacements of charges associated with integral membrane proteins. A technology was developed for incorporating membrane receptor proteins into matrices that mimic a natural lipid bilayer but which are bound to the surface of solid electrode materials. The focus of the investigation was on the photoreceptor protein, rhodopsin, as a model for a broad class of membrane receptor proteins.

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Dr. R.J. Nowak/Dr. I. Vodyanoy	(703) 696-4409/4056	ONR

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FINAL REPORT
Contract N00014-88-C-0193
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Principal Investigator: H. Gilbert Smith, Ph.D.

Contractor: TSI Mason Research Institute

Title: Surface-Bound Membrane-Mimetic Assemblies:
Electrostatic Attributes of Integral Membrane Proteins.

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Cognizant ONR Scientific Officer: Dr. R. J. Nowak / Dr. I. Vodyanoy

Description of Project

The objective of this project was to develop a system for investigating stimulus-induced displacements of charges associated with integral membrane proteins. A technology was developed for incorporating membrane receptor proteins into matrices that mimic a natural lipid bilayer but which are bound to the surface of solid electrode materials. The focus of the investigation was on the photoreceptor protein, rhodopsin, as a model for a broad class of membrane receptor proteins.

Summary of Results

The membrane reconstitution technique of detergent dialysis was adapted to assemble membrane structures onto solid surfaces that had been made partially hydrophobic through covalent attachment of long-chained alkyl groups. The alkyl groups attached to the surface appeared to provide an initiation point for membrane formation. The structures formed were characterized by ac impedance, FTIR and compositional analysis. The results were generally consistent with the formation of a lipid bilayer structure anchored to the surface by the surface-attached alkyl chains. The composition and electrical characteristics indicated a single membrane layer. This approach incorporated receptor proteins at greater than 10% of their density in native membranes. Rhodopsin incorporated into these structures retained its native optical absorption spectrum and preliminary results suggested that it also retained its native ability to activate a G-protein cascade that results in the hydrolysis of cGMP.

The approach developed in this project allows one to form a structure that mimics many of the properties of a biological membrane on any surface that can be modified by silanization. These structures can be formed in intimate contact with an electrode surface and thus have potential for monitoring charge movements in the low dielectric membrane core. In addition to such basic research applications, these biomimetic structures may also provide a means for incorporating receptor proteins into sensing devices. Details of the approach and the results obtained are given in various publications and presentations.

Publications

H. G. Smith, Surface-Bound Biomembrane Structures for use in Biosensors. Proceedings of the Symposium on Agents of Biological Origins, American Defense Preparedness Association and US Army CRDEC, Applied Physics Laboratory, Johns Hopkins University, March 1989, pp 44-52.

H. G. Smith, J. Li, N. W. Downer & L. W. DeLuca, Surface-Bound Biomembrane Assemblies. Proceedings of the IEEE/EMBS 11, 1329 (1989).

J. Li, N. W. Downer & H. G. Smith, Impedance Analysis of Surface-Bound Biomembranes. Proceedings of the IEEE/EMBS 12, 498 (1990).

J. Li, N. W. Downer & H. G. Smith, Evaluation of Surface-Bound Membranes with Electrochemical Impedance Spectroscopy. 1991. Submitted as a chapter for an ACS Advances in Chemistry volume on Membrane Electrochemistry. (1991)

N. W. Downer, J. Li, L. W. DeLuca, E. M. Penniman & H. G. Smith, Surface-Bound Alkyl Monolayers: Electrochemical and Structural Characterization. Submitted to Analytical Chemistry (1991).

N. W. Downer, J. Li, E. M. Penniman & H. G. Smith, Surface-Bound Biomembranes Incorporating Receptors: Electrochemical and Structural Characterization. Submitted to Biosensors & Bioelectronics (1991).

Invited Presentations

H. G. Smith, J. Li, N. W. Downer & L. W. DeLuca, "Surface-Bound Biomembrane Assemblies", 11th Annual International Conference of the IEEE Engineering in Medicine and Biology Society, Seattle, Washington, Nov. 9-12, 1989.

H. G. Smith, N. W. Downer & J. Li, "Surface-Bound, Biomembrane Assemblies Incorporating Receptor Proteins for Biosensor Applications", Symposium on Biosensors, SUNY Buffalo, Buffalo, NY, Nov. 13, 1989.

H. G. Smith, "Surface-Bound, Biomembrane Assemblies Incorporating Receptor Proteins for Biosensor Applications", Northeastern Section, American Chemical Society, Medicinal Chemistry Group, Boston College, Dec. 12, 1989.

J. Li, N. W. Downer & H. G. Smith, "Impedance Analysis of Surface-Bound Biomembranes", 12th Annual International Conference of the IEEE Engineering in Medicine and Biology Society, Philadelphia, PA, Nov. 1-4, 1990.

H. G. Smith, "Receptor Biosensors", Biomedical Engineering Departmental Seminar, Worcester Polytechnic Institute, Worcester, MA, March 11, 1991.

Other Presentations

H. G. Smith, "Surface-Bound Biomembrane Structures for use in Biosensors", Symposium on Agents of Biological Origins, American Defense Preparedness Association and US Army CRDEC, Applied Physics Laboratory, Johns Hopkins University, March 1989.

N. W. Downer, J. Li & H. G. Smith "Capacitive Devices Incorporating Integral Membrane Proteins", UCLA Symposia on Molecular and Cellular Biology of Biosensors and Bioprobes, Frisco, CO, Feb 3-8, 1990.

Personnel Participating on Project

Principal Investigator	H. Gilbert Smith, Ph.D.
Other Professional Staff	J. Li, Ph.D.
	Nancy W. Downer, Ph.D.
Technicians	Wendy W. Penniman, B.S.
	Leslie W. Deluca, B.S.